

1) How important is it to follow all procedures in a Histology laboratory especially in relation to the tissue samples sent for analysis?

- Tissues cannot be replaced
- Ensure accurate analysis
- Accept all samples without rejection
- Do a detailed investigation and rectify any discrepancies

2) a) Figure 1. Complete the table below by naming the components and state the function for each number



1	Universal cassette clamp	Holds the paraffin wax block in place for sectioning.
2	Knife holder	Holds the knife in place for trimming and sectioning
3	Knife holder base	Allows the knife to move sideways
4	Clamp lever for knife holder base	Lets the knife holder move back and forth for alignment.
5	Directional fixture for specimen clamp	Do not adjust. Changes the angle of the cassette during sectioning.
6	Handwheel handle with locking function	The handle also acts as a brake for the handwheel.
7	Handwheel	Moves forward (clockwise) to advance the cassette holder for sectioning
8	Specimen retraction ON/OFF	Stops the blade from touching the ribbon on the return stroke.
9	Handwheel brake lever	Acts as a brake for the handwheel.

b) Figure 2. Complete the table below by naming the components and state the function for each number



1	Course feed wheel	Moves the cutting head forward or backward to align the knife and block before cutting.
2	Course wheel running direction selector	Changes the direction of the coarse wheel for advancing and retracting.
3	Mechanical trimming function lever	First stop at 10µm and second stop at 50µm per wheel turn for quick trimming.
4	Section thickness selector knob	Sets the desired thickness of final sections.
5	Section thickness display window	Thickness in micrometers (µm)

3) a) Complete the table below for **Tissue Processing**. Detail what reagents are used in each step and the purpose of each step.

Fixation	Reagents used:
	Neutral buffered formalin
	Purpose of the step: Fixes the tissue to prevent decay and preserve its structure by crosslinking proteins.
Dehydration	Reagents used:
	70-100% Ethanol's with at least 3x 100% Ethanol.
	Purpose of the step: Removes water from the tissue to prepare it for the clearing reagent.
Clearing	Reagents used:
	Xylene or Xylene substitute
	Purpose of the step: Removes the ethanol from the tissue to allow the paraffin to infiltrate.
Infiltration	Reagents used:
	Paraffin wax at least 3x Paraffin baths
	Purpose of the step: Permanently preserves the tissue, hardens it for sectioning, and provides support.

b) Complete the table below for **Tissue Staining: Haematoxylin and Eosin**. Detail what reagents are used in each step and the purpose of each step.

Dewax	Reagents used:
	Xylene or Xylene substitute
	Purpose of the step: Removes wax from the section for staining
Rehydration	Reagents used:
	At least 2x 100% Ethanol followed by 90-70% ethanols
	Purpose of the step: Replaces xylene and rehydrates tissue for staining.
Staining (Stain 1)	Reagents used:
	Harris's Haematoxylin
	Purpose of the step: Stains nuclei, cartilage, and mucins
Differentiation	Reagents used:
	Acid alcohol
	Purpose of the step: Removes the excess Haematoxylin stain
Bluing	Reagents used:
	Scott's Tap Water
	Purpose of the step: Changes the Haematoxylin stain from red to blue

Continue

Counter Staining (Stain 2)	Reagents used:
	1% Alcoholic Eosin
	Purpose of the step: Stains non-nuclear elements in shades of pink and orange.
Dehydrate	Reagents used:
	90-100% Ethanol with at least 2x 100% Ethanol.
	Purpose of the step: Removes water from the tissue before permanent mounting
Clearing	Reagents used:
	Xylene at least 2x Xylene
	Purpose of the step: Removes ethanol from the tissue to allow DPX mounting.
Mounting (DPX)	Reagents used:
	DPX (Di-Polystyrene Xylene)
	Purpose of the step: Permanently preserves the tissue section and allows for viewing. It has the same refractive index as glass.

4) a) You have received a tissue sample in a 70mL Formalin pot, and it has been found to be leaking, what steps do you need to go through to rectify the

situation? And does this incident require further follow up e.g. a hazard or incident report. Please explain your answer?

- Put a paper towel under the leaking container to catch drips.
- Take the specimen container to a sink.
- Pour the aldehyde solution into a hazardous waste container.
- Clean any contaminated surfaces with cold water twice.
- Put all contaminated paper towels in a sealable bag for hazardous waste disposal.
- Follow up: None needed – minor incident.
- Follow up? Nothing – minor incident

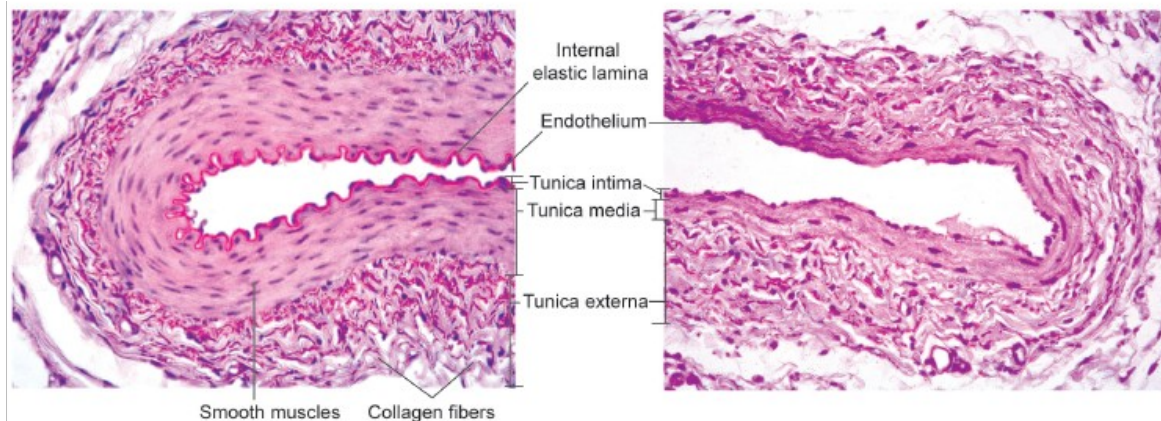
b) A Formalin storage tank of 20L was knocked over and its contents spilled over the floor of the reagent room, what steps do you need to go through to rectify the situation? And does this incident require further follow up e.g. a hazard or incident report. Please explain your answer?

- Contain the spill if possible.
- Wear PPE: nitrile gloves, disposable gown or overalls, waterproof footwear or shoe covers, and a full-face respirator or self-contained breathing apparatus.
- Encircle the spill with absorbent material and mix it in from the outside.
- Wait for Emergency Response Personnel.
- If safe, use absorbent spill control material to contain the spill. Preferably use a formalin absorbent and neutralizer like Spilfyter, but general chemical absorbents, pillows, or bunds can also be used.
- Clean-up should be done with Emergency Response Personnel.
- Follow up? both incident and hazard report – this is a major spill

c) In the reagent preparation room, the fume hood has failed and no longer is running what steps do you need to go through to rectify the situation? And does this incident require further follow up e.g. a hazard or incident report. Please explain your answer?

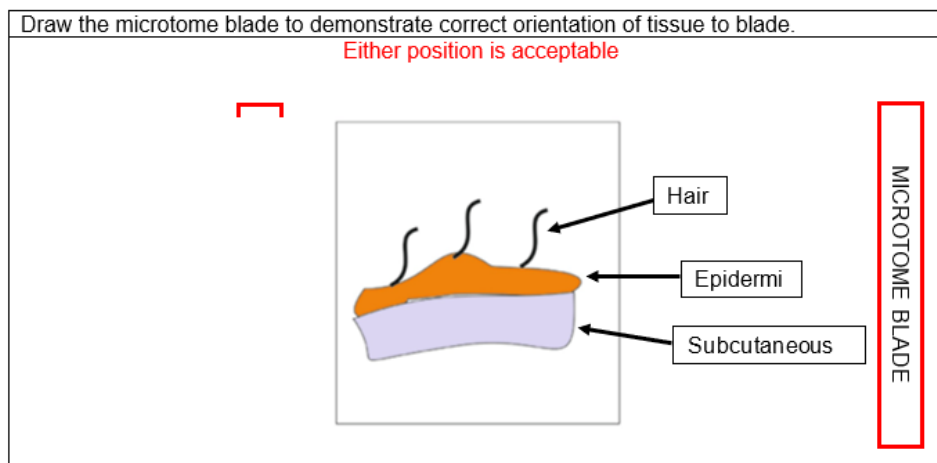
- Remove all reagents and equipment from the fume hood.
- Alert nearby staff about the issue.
- Attach a “DO NOT OPERATE” tag to the fume hood.
- Notify your supervisor.
- Follow up? an incident form. – this is a major incident

- 5) For the following photos Figure 3 of blood vessels identify the following:
Which photo is an artery or vein?
How the staining of the different layers helps to identify the tissue?



What type of Vessel	
artery	vein
Staining and Identifying features	
- Thick tunica media with up to 40 layers of smooth muscle. - Thicker endothelium layer with prominent staining. - Thick tunica externa with connective tissue.	- Only 8 layers of smooth muscle in the tunica media. - Thinner tunica externa. - Less prominent endothelium.

- 6) a) For the following photo of a skin section in a wax block draw and label where the microtome knife should be located.



- b) Why is it essential to have the correct orientation of the skin layers to the microtome blade? What steps do you need to take to produce good quality sections?

It is important to orientate the tissue parallel to the blade to ensure no compaction occurs during sectioning.

Steps:

- Orient the tissue parallel to the blade to avoid compaction.
- Place the tissue in the wax block with the cassette label on the right.
- Chill the tissue for at least 20 minutes to harden the wax and tissue.
- Maintain an even and consistent sectioning speed for uniform tissue thickness.
- Trim the full face before sectioning.

7) In a busy Histology laboratory that processes over 300 blocks per day. The tissue processor is on a once per fortnight reagent changing schedule. Describe what effect the reagent schedule may have on the processor reagents and the subsequent effects it may have on embedding, microtomy and staining?

- The reagents are of poor quality.
- Reagents may not fully remove ethanol and xylene from the paraffin blocks.
- Excess ethanol or xylene in the wax block can cause:
- Poor embedding
- Poor sectioning
- Poor staining

8) Complete the following table: For each listed hazard relate it to the area of the Histology laboratory process (e.g. Specimen receipt, Grossing, Processing, Embedding, Microtomy and Staining) and briefly describe the safety procedure or precaution for each identified hazard.

Hazard	Area in the Laboratory	Procedure/Precaution
Formaldehyde (Formalin)	Specimen reception Grossing Processing	Work in a well ventilated area with VOC extraction Down draft work benches PPE Have formalin spill kits available.
Ethanol	Processing Staining	PPE Use fume hoods with active carbon filters on processors
Xylene	Processing Staining Microtomy	PPE Use fume hoods with active carbon filters on processors Use Xylene substitute
Repetitive Strain Injury (RSI)	Embedding Microtomy	Take regular breaks every 20 mins Purchase ergonomic or motorized machines
Sharps (Not glass)	Grossing Microtomy	Use knife covers and remove knives before working on the block. Apply wheel brakes when changing blocks or knives. Plastic disposable scalpel.
Glass	Microtomy Staining	PPE Forceps Caution Use plastic staining containers Coplin jars.

- 9) Complete the following table: For each work area of the Histology laboratory process (e.g. Grossing, Processing, Embedding, Microtomy and Staining) and briefly describe one (1) sustainability measure.

Work Area	Sustainability Measure
Grossing	Follow the correct SOP Use a formalin recycler
Processing	Implement reagent management system Use solvent (Ethanol and Xylene) recyclers
Embedding	Follow the correct SOP Choose the appropriate mold size
Microtomy	Use old sectioning blades to trim blocks Follow the correct SOP
Staining	Solvent recyclers Wipe excess stain from slides to reduce carryover and the need for reagent replacement

- 10) a) Explain In detail what traceability steps are taken in the Histology laboratory in relation to samples and cut tissue? (think about every step of the process e.g. sample reception, grossing, processing, embedding, microtomy and staining)

- Each sample gets a unique lab number at reception.
- During grossing, the sample is described, and a block key is created, noting the number of slices in each cassette. A ticket with the lab number, tissue type, block number, and number of pieces is placed in each cassette.
- At embedding, the ticket is checked for the lab number and the number of pieces in the cassette.
- At microtomy, the number of pieces is checked again to ensure proper trimming.
- At staining, a final check ensures the block, slides, and tissue pieces match before reporting.

- b) What laws, legislation and accrediting bodies regulations cover Confidentiality for the Pathology industry? Name at least **FOUR (4)**

ISO 17025, Good Laboratory Practices,
Privacy Act 1988,
Freedom of Information Act 1992,
NPAAC, Health service act 2016
My health Act 2012
Health Practitioner Regulation National Law (WA) act 2010
Australian Commission on Safety and Quality in Health Care (ACSQHC)
The Royal College of Pathologist of Australasia (RCPA)

- c) What Federal body and State Government act cover the Retention and Disposal of Patient records? State **both**
NPAAC

WA States Records Act 2000 Patient Information Retention and Disposal Schedule

d) The NPAAC “Retention of Laboratory Records and Diagnostic Material” require Histology laboratories to retain human tissue for minimum retention times. Give **two examples** of the material held and the correct retention time for both examples?

- Slide 10 years
- FISH 6 months
- Blocks 10 years
- Frozen Section 10 years
- No residual tissue sample 1 month
- Tissue in reserve tissue 1 month

e) The NPAAC “Requirements for The Performance of Anatomical Pathology Cut-UP” require Histology laboratories define who can perform tissue grossing. State who the ultimate person/s is in relation to performing, delegating grossing responsibilities and troubleshooting grossing uncertainties?

Pathologist

f) ISO 17025 requires laboratories to implement procedures to ensure the security of patient records. State **ONE (1)** such requirement.

- **Have a documented process to ensure all employees know and understand the confidentiality policy, including how to monitor and update it.**
- **Implement procedures to prevent former employees from using or disclosing confidential information without permission after they leave.**
- **Create a written information security policy that outlines the lab’s confidentiality obligations and practices to protect confidential information.**
- **Ensure confidential information is well protected within the lab.**
- **Store confidential paper documents and records in a secure location, away from non-lab personnel.**
- **Shred confidential paper documents when no longer needed.**
- **Store electronic documents on a secure network and view them only on secure devices.**
- **Share information with other personnel only when necessary and authorized.**

g) NPAAC “Requirements for Medical Pathology Services – Section 2” outline Ethical practices. State **ONE (1)** such requirement.

- The wellbeing and rights of patients must be prioritised.
- Medical Pathology Services should have an open disclosure policy to discuss any patient safety incidents that caused or could have caused harm.
- Patients, their specimens, and body parts must be treated with respect.
- Ethical standards must be defined through policies and procedures.
- There must be a policy on informed consent that meets jurisdictional requirements.

- Patients' personal information must always be kept private and confidential.